DFW

Patent
Attorney Docket No. 82325
Customer No. 23685

TRANSMITTAL LETTER

Inventors: Reimo Tetzner et al.
Serial No: 10/585,682
Filed: 7-10-06
Group Art Unit:
Examiner: Unknown
Batch No:
Notice of Allowance:
For: METHOD FOR INVESTIGATING CYTOSINE METHYLATION IN DNA BY MEANS OF DNA REPAIR ENZYMES

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

Transmitted herewith for the above-identified patent application are the following:

An English Translation of the International Preliminary Report on Patentability
A return postcard

The item(s) checked below are appropriate:

1. __ Applicant(s) hereby petition(s) for a ( ) month extension of time to respond to an dated .

2. X Please charge any fees or costs not accounted for to Deposit Account No. 11-1755.

Date: November 6, 2006

Edward M. Kriegsman
Reg. No. 33,529

KRIEGSMAN & KRIEGSMAN
30 Turnpike Road, Suite 9
Southborough, MA 01772
(508) 481-3500

I hereby certify that this correspondence is being deposited with the United States Postal Service as first class mail in an envelope addressed to: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450 on November 6, 2006

Edward M. Kriegsman

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY
(Chapter I of the Patent Cooperation Treaty)

(PCT Rule 44*bis*)

Applicant's or agent's file reference P1075PC00	**FOR FURTHER ACTION**	See item 4 below
International application No. PCT/EP2005/000231	International filing date *(day/month/year)* 10 January 2005 (10.01.2005)	Priority date *(day/month/year)* 09 January 2004 (09.01.2004)
International Patent Classification (8th edition unless older edition indicated) See relevant information in Form PCT/ISA/237		
Applicant EPIGENOMICS AG		

1. This international preliminary report on patentability (Chapter I) is issued by the International Bureau on behalf of the International Searching Authority under Rule 44 *bis*.1(a).

2. This REPORT consists of a total of 7 sheets, including this cover sheet.

   In the attached sheets, any reference to the written opinion of the International Searching Authority should be read as a reference to the international preliminary report on patentability (Chapter I) instead.

3. This report contains indications relating to the following items:

☒	Box No. I	Basis of the report
☒	Box No. II	Priority
☐	Box No. III	Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
☐	Box No. IV	Lack of unity of invention
☒	Box No. V	Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
☐	Box No. VI	Certain documents cited
☒	Box No. VII	Certain defects in the international application
☒	Box No. VIII	Certain observations on the international application

4. The International Bureau will communicate this report to designated Offices in accordance with Rules 44*bis*.3(c) and 93*bis*.1 but not, except where the applicant makes an express request under Article 23(2), before the expiration of 30 months from the priority date (Rule 44*bis* .2).

	Date of issuance of this report 10 July 2006 (10.07.2006)
The International Bureau of WIPO 34, chemin des Colombettes 1211 Geneva 20, Switzerland Facsimile No. +41 22 338 82 70	Authorized officer Ellen Moyse e-mail: pt05@wipo.int

Form PCT/IB/373 (January 2004)

PATENT COOPERATION TREATY

From the
INTERNATIONAL SEARCHING AUTHORITY

To:

see form PCT/ISA/220

PCT REC'D 19 MAY 2005
WIPO PCT

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY
(PCT Rule 43*bis*.1)

Date of mailing
(day/month/year) see form PCT/ISA/210 (second sheet)

Applicant's or agent's file reference
see form PCT/ISA/220

FOR FURTHER ACTION
See paragraph 2 below

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/EP2005/000231	10.01.2005	09.01.2004

International Patent Classification (IPC) or both national classification and IPC
C12Q1/68

Applicant
EPIGENOMICS AG

1. This opinion contains indications relating to the following items:

- ☒ Box No. I Basis of the opinion
- ☒ Box No. II Priority
- ☐ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☐ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- ☐ Box No. VI Certain documents cited
- ☒ Box No. VII Certain defects in the international application
- ☒ Box No. VIII Certain observations on the international application

2. **FURTHER ACTION**

If a demand for international preliminary examination is made, this opinion will usually be considered to be a written opinion of the International Preliminary Examining Authority ("IPEA"). However, this does not apply where the applicant chooses an Authority other than this one to be the IPEA and the chosen IPEA has notifed the International Bureau under Rule 66.1*bis*(b) that written opinions of this International Searching Authority will not be so considered.

If this opinion is, as provided above, considered to be a written opinion of the IPEA, the applicant is invited to submit to the IPEA a written reply together, where appropriate, with amendments, before the expiration of three months from the date of mailing of Form PCT/ISA/220 or before the expiration of 22 months from the priority date, whichever expires later.

For further options, see Form PCT/ISA/220.

3. For further details, see notes to Form PCT/ISA/220.

Name and mailing address of the ISA:

European Patent Office
D-80298 Munich
Tel. +49 89 2399 - 0 Tx: 523656 epmu d
Fax: +49 89 2399 - 4465

Authorized Officer

Hennard, C

Telephone No. +49 89 2399-7355

Box No. I Basis of the opinion

1. With regard to the **language**, this opinion has been established on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.

   ☐ This opinion has been established on the basis of a translation from the original language into the following language , which is the language of a translation furnished for the purposes of international search (under Rules 12.3 and 23.1(b)).

2. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application and necessary to the claimed invention, this opinion has been established on the basis of:

   a. type of material:

   ☒ a sequence listing

   ☐ table(s) related to the sequence listing

   b. format of material:

   ☒ in written format

   ☒ in computer readable form

   c. time of filing/furnishing:

   ☒ contained in the international application as filed.

   ☒ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority for the purposes of search.

3. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

4. Additional comments:

Box No. II Priority

1. ☒ The validity of the priority claim has not been considered because the International Searching Authority does not have in its possession a copy of the earlier application whose priority has been claimed or, where required, a translation of that earlier application. This opinion has nevertheless been established on the assumption that the relevant date (Rules 43*bis*.1 and 64.1) is the claimed priority date.

2. ☐ This opinion has been established as if no priority had been claimed due to the fact that the priority claim has been found invalid (Rules 43*bis*.1 and 64.1). Thus for the purposes of this opinion, the international filing date indicated above is considered to be the relevant date.

3. Additional observations, if necessary:

Box No. V Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	14-15, 19-20
	No:	Claims	1-13, 16-18, 21-23
Inventive step (IS)	Yes:	Claims	None
	No:	Claims	1-23
Industrial applicability (IA)	Yes:	Claims	1-23
	No:	Claims	None

2. Citations and explanations

see separate sheet

Box No. VII Certain defects in the international application

The following defects in the form or contents of the international application have been noted:

see separate sheet

Box No. VIII Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

see separate sheet

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Reference is made to the following documents:
   D1: DE 198 53 398 C1
   D2: DE 102 04 566 A1
   D3: JOURNAL OF BIOLOGICAL CHEMISTRY, vol. 277, no. 42, 18 October 2002, pages 39926-39936,
   D4: ELECTROPHORESIS, vol. 20, no. 6, June 1999 (1999-06), pages 1141-1148,
   D5: HUMAN MUTATION, vol. 20, no. 2, 2002, pages 139-147,
   D6: US-A-5 656 430

2. **Novelty (Article 33(2) PCT):**

   2.1 **D1** (column 3, line 25 - column 4, line 30; claims 1, 3, 10) describes a method for analysing the cytosine methylation in a DNA comprising the steps of chemically converting the non-methylated cytosine into uracil using the bisulphite conversion reaction. The obtained DNA is amplified and hybridized with a complementary strand forming an hetero duplex. The duplex is cleaved in mismatch position using an enzymatic reaction (involving MutH, MutL or MutS repair proteins) and the cleaved DNA is analysed using preferably mass spectroscopy. The hybridization is preferably performed on an solid support. An example further describes the method involving 97 different sources of cellular DNA in which each sample is compared to a reference. In the light of **D1, claims 1-12, 16-18** are not new.

   2.2 **D2** (page 2, lines 29-55 and 60-65; page 3, lines 35-40; claim 1) discloses a method for determining the methylated cytosine in a DNA involving the bisulphite reaction conversion of cytosine into uracil followed by hybridization of the modified DNA with another strand forming mismatch pairing and reacting the hetero duplex DNA with enzymes among which MutY is cited. Since MutY is one of the most preferred enzymes used in the application and since it is a DNA repair enzyme which cleaves the DNA in mismatch positions, the cleavage of the DNA strand in the mismatch position by the enzyme is implicit. In the light of **D2, claims 1-6, 11-13, 16-18** are not new.

   2.3 **D3** (page 39928, last paragraph) describes a process for monitoring the glycosylase steps in the BER-assays which involves the use of the mismatch repair enzyme in the presence of an hetero duplex presenting U:G mismatch.

The duplex is amplified using a polymerase as described in the BER assay of the document. This disclosure is considered to anticipate the kits of **claims 21-23** of the present application.

2.4 **D4** (page 1145, paragraph 3.3), **D5** (page 141, middle paragraph, left-hand column) and **D6** (example 1, claims) disclose a process involving a T/G mismatch duplex repaired by a thermostable TDG enzyme or MutY repair enzyme. These documents are anticipating the kit of **claims 21-22** of the present application.

2.5 In order to summarise the above objections, **claims 1-13, 16-18 and 21-23** of the present application are not novel and do not fulfil the requirements of **Article 33(2) PCT** whereas **claims 14-15 and 19-20** are novel.

3. **Inventive merit (Article 33(3) PCT):**

D1 (see passages above), which is the closest prior art, concerns a method for determining the methylation state of cytosines in a DNA. The process of **claim 14** of the present application distinguishes itself from **D1** by the use of a heat stable enzyme during the enzymatic cleavage of the mismatch DNA.

No technical effect is achieved by the use of the specific enzyme, thus the problem to be solved can be seen as the provision of an alternative to the method of **D1** which does not specify the nature of the enzyme.

D4 teaches that the thermostable enzyme TDG is suitable for the cleavage of mismatch DNA. Thus, the skilled person in charge of providing an alternative enzyme to the method of **D1** would consider the teaching of **D4** and use a TDG enzyme in a method as in **D1** without demonstrating an inventive merit. Therefore, **claims 14 and 15** of the present application do not involve an inventive merit over the combination of **D1** with **D4**.

Further, the preferred embodiments of **claims 19 and 20** relate only to the origine of the DNA sample to be tested according the method of the application. Such feature does not involve an inventive merit because it is considered as standard modification in the field of selecting specific samples to perform the method.

It is concluded that **claims 14-15 and 19-20** do not involve an inventive merit and do not fulfil the requirements of **Article 33(3) PCT**.

4. **Industrial applicability (Article 33(4) PCT):**

An industrial applicability of the invention is obvious and **claims 1-23** of the present application are considered to fulfil the requirements of **Article 33(4) PCT**.

Re Item VII

Certain defects in the international application

5. Contrary to the requirements of **Rule 5.1(a)(ii) PCT**, the relevant background art disclosed in **D1-D2** is not mentioned in the description, nor are these documents identified therein.

Re Item VIII

Certain observations on the international application

6. In step b) of **claim 1 and in claim 3** of the present application, the hybridisation is characterised as not forming hybrids when a specific methylation status occurs. Since methylcytosine has the same hybridization properties as cytosine and since the conversion of unmethylated cytosine into uracil leads to a mismatch, it is not clear which methylation status is supposed not to form a hybrid. From the description on page 7, it is mentioned that the stringency conditions are selected such that either mismatch or no hybrid occurs but no mention could be found that one methylation status forms a mismatch and the other no hybrid. Thus **claims 1 and 3** are considered unclear and not supported by the description and do not fulfil the requirements of **Article 6 PCT**.